PME RESEARCH LABORATORY, SEPTEMBER 1980

PROJECT TITLE : PROTAGORAS

PERIOD COVERED : SEPTEMBER 1 - 30, 1980

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The objective of the project PROTAGORAS is to produce cigarettes of tobacco from which protein has been removed. This is in order to eliminate some of the precursors of nitrogen-containing smoke constituents. At a later stage of the project, specific protein fractions will be re-added to the deproteinated filler for taste improvement.

1. ACTIVITY OF PROTEOLYTIC ENZYMES (1)

The Azocoll test (2) was used to determine the activity of proteolytic enzymes currently applied in our trials. One unit represents the activity that releases enough Azo dye to give an absorption of 0.001 per min. under test conditions.

Enzyme	Manufacturer	Units/mg
Protease VII	Sigma	9966
Protease VIII	"	12322
Protease X	"	10416
Pronase	Boehringer	2216
Protease	Calbiochem	10077

2. EXTRACTION OF PROTEIN FROM THE B BLEND USING DIFFERENT PROTEASES (3)

Conditions: Tobacco : water = 1 : 10
pH = 7.5
Agitation = 140 r.p.m.
Incubation time = 6 hrs.
Enzymes = 375 mg/100 g tobacco.

Enzyme	% Protein extracted at	
	37° C	50° C
Control	34	35
Protease VII	58	53
Protease VIII	61	55
Protease X	51	53
Pronase	57	50

Extractions worked better at 37° C than at 50° C. No obvious correlation exists between the activity of the enzymes and the amount of protein extracted.

3. ELIMINTATION OF PROTEIN FROM TOBACCO EXTRACT (4)

Tobacco extracts from the above described extraction trials were passed through an Amicon ultrafiltration unit. Following protein eliminations could be obtained:

Minimal Molecular Weight of Retained Protein	Elimination of Protein %
5000	21
10000	14-16
50000	6

The results show that proteins extracted from tobacco with proteolytic enzymes are rather small. Ultrafiltration cannot be the only step for their elimination.

REFERENCES

(1) A. Haenggi, Notebook 791201, p. 1-7.

(2) Calbiochem-Behring Corp., Internal Publication on Azocoll, 1979.

(3) A. Haenggi, Notebook 791201, p. 8-12, p. 14-15.

(4) A. Haenggi, Notebook 791201, p. 13.

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